August 31, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 4793-review.doc).

Title: Reduced hemoglobin and increased C-reactive protein are associated with upper gastrointestinal bleeding

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Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript: NO: 4793

The manuscript has been improved according to the suggestions of reviewers:

Manuscript Tomizawa M. et al. entitled “Reduced hemoglobin and increased C-reactive protein as markers of upper gastrointestinal bleeding” is devoted to retrospectively analysis of upper gastrointestinal (GI) bleeding and search predictors of bleeding. The title reflects the content of the article. The abstract structured and contains sections of the aim, methods, main results and conclusion. The authors in abstract of manuscript present the aim of the research as the conclusion. The introduction short and contains information on the role of endoscopy in the diagnosis and treatment of bleeding from the upper gastrointestinal tract. The design of the study is rational.

Materials and methods do not contain a description of endoscopy method.

Response: Endoscopy methods were added in the beginning of Materials and methods.

Clinical characteristics of patients are practically absent. Laboratory analysis isn't cited in this part.

Response: Table 2 was added to present laboratory data.

The results. The figures, tables demonstrate the results and contain statistical materials. Submitted references aren't cited papers from WJG. The study is of particular interest to the practical medicine.

Response: Articles in World Journal of Gastrotenterology by Ljubicic et al. and Leffroy et al. were cited.

1. You indicate there were 1101 endoscopies but only 156 patients enrolled (14%) and only 17 had UGI bleeding (1.5%. What did the other patients have? How was the decision to perform endoscopy made? Why were so few patients enrolled in the study?

Response: 1101 were total endoscopy during this period. Indications of 1101 endoscopies were screening, anemia, examination of abdominal symptoms and other reasons. We chose patients who had laboratory data around the day of endoscopy and three months before endoscopy to analyze reduction rate. Patient number, thus, reduced to 156 patients. 17 patients with bleeding matched the criteria and were analyzed. The other patients were excluded from the present study. This part was added in the second paragraph of Materials and Methods.

2. How did you discriminate upper from lower GI bleeding? Do the parameters you utilized also work in lower GI bleeding?

Response: It is hard to discriminate upper GI bleeding from lower GI bleeding with the parameters we presented. Upper endoscopy and colonoscopy should be performed for patients with low Hb {Bull-Henry, 2013 #2614}. Our hospital followed this theory. Low Hb include patients with upper or lower gastrointestinal bleeding. Parameters analyzed in our present study were expected to be applied to lower gastrointestinal bleeding. This

part was added as the second paragraph of Discussion.

3. Why were patients getting laboratory studies performed 3 months before they had a bleed? is this routine? Usually patients present with bleeding and one does not have the luxury of checking labs 3 months before the bleed. Explain in detail how this occurred.

Response: We chose patients who had laboratory data three months before endoscopy. Patients who did not have laboratory data three months before endoscopy were excluded from this study. Some patients were referred from other clinics or hospitals due to anemia. Others visited our hospitals after intervals more than six months. Patients who did not have laboratory data three months before endoscopy were not included in the present study. These were the reason why the patients analyzed in this study had laboratory data three months before endoscopy. This part was added in the second paragraph of Materials and Methods.

4. Of course, a low HCT suggests there is bleeding, but it does not discriminate the source of bleeding.How did you know there was upper Gi vs. lower GI bleeding in your patients? Did patients get both upper and lower endoscopy?

Response: It was hard to discriminate upper or lower gastrointestinal bleeding with a low HCT. As a reference recommended, upper endoscopy were performed in our hospital. If upper endoscopy was normal, patients were subjected to colonoscopy. This part was added as the second paragraph of Discussion.

5. Did any of the patients have any hereditary anemias that might have contributed to a low HCT? Thalessemia, sickle cell, etc. How were patients screened to eliminate these patients from the analysis?

Response: Patients with anemia were investigated following Goddard et al (BS39). Upper endoscopy was performed. If no abnormal findings were absent, they were subjected to colonoscopy. When no abnormal findings were found, they would be investigated for causes of anemia, such as thalessemia and sickle cell anemia (BQ12). This part was added as the second paragraph

of Discussin. In our study, no patients were with thalessemia or sickle cell anemia. This part was added in the second paragraph of Materials and Methods.

This manuscript attempted to establish biomarkers for upper gastrointestinal bleeding by measuring blood hemoglobin and C-reactive protein at 3 months apart. This work, however carefully conducted, failed to establish the clinical relevancy for the following reasons:

1. Normal values of hemoglobin and C-reactive protein already existed. A patient appeared for have low hemoglobin and high C-reactive protein should not be asked to wait for 3 months before clinical action. Combining with other symptoms of upper GI bleeding an experienced GI clinician should be able to identify, endoscopy and treatment should be initiated immediately. As shown in Table 1, one of the causes of upper GI bleeding is gastric cancer. In that case, delaying clinical action is especially undesirable.

Response:
Mahlknecht et al report age-related change of Hb {Mahlknecht, 2010 #2613}. They excluded patients who had hematological history, oncological diseases, chronic infection or inflammation. They show that Hb decreases as aging, and that average Hb is, interestingly, below normal range of Hb. When patients with low Hb are encountered, Hb of previous days is always checked. Decreasae of Hb suggested that the patients had bleeding diseases, such as gastric ulcers. Therefore, we analyzed the change rate of Hb and other paramters. This part was added as the third paragraph of Introduction.

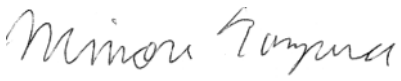
2. Blood loss from any part of the body will lead to low hemoglobin. C-reactive protein is a non-specific indicator of inflammatory response. Thus, many disease conditions can lead to low hemoglobin and high C-reactive protein. These two measurements do not have the specificity to serve as biomarkers for a general practitioner. It is possible for the work to be published as "Reduced hemoglobin and increased C-reactive protein

ARE ASSOCIATED WITH upper gastrointestinal bleeding" after revision. This modified title more accurately reflects the limited contribution of the work.

Response: Title was changed to "Reduced hemoglobin and increased C-reactive protein are associated with upper gastrointestinal bleeding"

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,



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